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The Effects of Cinnamon on Newcastle Disease Performance and Hemato-Antibodies Titer in Broilers Iraq

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Abstract : This study will look into the effects of dietary cinnamon supplementation. Immunological serum tests are used to boost broiler immunity to the New Castle vaccine. Ross' flock consists of 308 one-day-old broiler chicks. T1 chicks received 2% cinnamon powder in their feed, T2 chicks received 4% cinnamon powder in their feed, and T3 chicks received 300 ppm cinnamon extract in their drinking water. Water and food must be provided for the duration of the thirty-day experiment. The first and second doses of the New Castle vaccine were administered to the birds. Titers of hem agglutination inhibition antibodies and antibody titers in an ELISA test were determined for these immunizations. Following the first and second vaccines, blood parameters such as the number of white blood cells and the heterophil/lymphocyt (H/L) ratio were also measured.

Hem agglutination inhibition antibodies and New Castle vaccination titers increased significantly in (T1, T2, T3) when compared to the control group (P 0.05). The ELISA test results showed a significant increase in New Castle vaccine titer antibodies (P 0.05). There were no significant differences in ELISA antibody titer or HI between the three groups treated with cinnamon extract at doses of 2%, 4%, and 300 ppm, respectively. In all groups, there was a significant increase in the standard of antibodies measured by the HI and ELIZA tests after the second vaccine compared to the first vaccine. The rate of white blood cells increased significantly (P 0.05) in the treatment groups when compared to the negative control group.

A study on the ratio of heterophils to lymphocytes was also conducted, and an increase in this ratio indicates stress. After the first and second vaccinations, there was a significant (P 0.05) decrease in the percentage of H/L cells in the treated groups compared to the control group, which reflected positively on the birds' health

Key words: Newcastle disease, Cinnamon, ELISA, HI., WBC Count.

Introduction

Newcastle disease is one of the diseases of great economic importance to poultry because it causes enormous economic losses that have a significant impact on the Iraqi economy, so one of the important matters is the process of conducting vaccinations and raising immunity through the use of some plant extracts after giving vaccines, and these plants are (Cinnamon plant) medicinal plants, and their extracts play a significant role (1) (2). Medicinal plants as a whole, plant extracts, and/or essential oils are combined or used separately in the diet. Cinnamon belongs to the Lauracea family. Cinnamon is a medicinal plant that has anti-oxidant, anti-diabetic, anti-septic, and local anesthetic properties. Many studies have been conducted to investigate the effectiveness of plants, plant extracts, and oils as antibiotic substitutes. In poultry diets, herbs and herbal products are used to replace synthetic and chemical ingredients (3). Cinnamon oil is widely used in the animal and poultry feed industries due to its distinct aroma (4). Cinnamaldehyde has the strongest antibacterial, antifungal, and antioxidant properties in cinnamon oil, followed by eugenol (5). Cinnamon oil may be a natural alternative to dietary antibiotics and may improve broiler chicken growth performance, according to some studies (6). However, no statistically significant improvement in chicken growth was found in other studies (7).

Materials and methods

In this experiment, 100 one-day-old Bulgarian-bred Ross (308) broiler chicks were used. The chicks were divided into four equal groups at random for this experiment and placed in specially prepared wooden cages. The first cage contained 24 chicks (control or comparison group C), the second cage contained 24 chicks

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(first treatment group T1), the third cage contained 24 chicks (second treatment group T2), and the fourth cage contained 24 chicks (control or comparison group T3) (control or comparison group T3). Heat, food, water, and aeration are all necessary for chicks to thrive. Cinnamon extract treatment groups to feed for the first, second, and third treatment groups (T1, T2, and T3) were added as a food additive to the first and final diets without any medical antibacterial concentrations of 2%, 4%, and 300 ppm cinnamon extract treatment groups, respectively (8). The standard diet was fed to the control group (C). After being vaccinated with the Newcastle vaccine for the duration of the trial, four unvaccinated chicks were used to conduct the hem agglutination test to determine the vaccination standard. The chicks were given the first Newcastle vaccination (B1) when they were one day old, and the second Lasota strain when they were 15 days old, both through drinking water. Blood was collected 14 days after the first immunization and 14 days after the second vaccination in anticoagulant-containing and anticoagulant-free tubes. Storage of serum at -18°C for immunological and serological testing.

Swabs of blood were taken directly from birds on glass slides by placing a drop of blood from a capillary tube on the slide, then spreading the blood with another glass slide on which a drop of blood is placed and pulled over the first slide at a 45-degree angle without applying pressure. After the blood dried, the slides were stained with a mixture of two Wright-Giemsa dyes according to the (9) method, and the percentage of H/L was calculated using a light microscope and a magnification of x 100 by placing a drop of oil on the slide according to the method (10).

WBC Count (Cell / mm^3)

Each treatment's blood was drawn from the brachial vein medial to the wing and placed in tubes containing the anticoagulant EDTA.

The counting method employed the Neubaur-Champer Haemocytometer (SUPE-RIOR, W. Germany), and a special Natt and Herrick's solution was used to dilute these cells in the blood of birds (11).

preparing for the counting process:

To count the white blood cells, a special pipette was used, and blood was drawn to the 0.5 mark, then the volume was completed to the 11th mark using the mentioned dilution solution, and the blood was thus diluted 20 times. Count the white blood cells in all of these boxes, as they appear dark blue and may be granular in shape. The total cell count was determined using the following equation:

The number of white blood cells in 1 ml of blood= (N / 4) × 20 × 10

N = the total number of white blood cells in the four squares.

4 = the number of large squares that the count was inside.

20 = number of times of dilution.

10 = used to get the total number in 1 mm ^{3} of blood, since the volume of blood in each of the four squares was 0.1 0 ml ^{3}

The results were statistically analyzed, which included one and two-way ANOVA, followed by a least significant test (P 0.05) to determine whether there were any significant differences between treatments (12).

Results

After the initial vaccination, the statistical analysis of the volumetric criterion of anti-Nuclear anti-agglutinating Newcastle antibodies revealed significant changes, with the treatment groups (T1, T2, and T3) outperforming the control group (C) with a significant difference (P 0.05).

The three treated groups (T1, T2, and T3) were significantly superior to the control group after the second vaccination (P 0.05), and the four groups were significantly superior in inhibitory Newcastle antibodies for hematopoiesis after the second vaccination over the first vaccination.

Table (1) Newcastle agglutinin antibodies that inhibit volumetric agglutination after first and second vaccination.

group	standard antibodies Log ^{10} First vac.	standard antibodies Log ^{10} Second vac.
C	2.2 ± 0.17 ±Bb	3.42 ± 0.12 Ba
T1	2.82 ± 0.32 ±Ab	4.25 ± 0.13 Aa
T2	2.81 ± 0.9 ±Ab	4.73 ± 0.17 Aa
T3	2.79 ± 0.8 ±Ab	4.51 ± 0.18 Aa

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* The numbers represent averages and standard deviations. * Capital letters represent the vertical statistical reading (between totals), while lowercase letters represent the horizontal statistical reading (between times). The similar letters indicate that no statistically significant differences exist between the transactions, whereas the different letters indicate that significant differences exist at the level of probability. (P < 0.05).

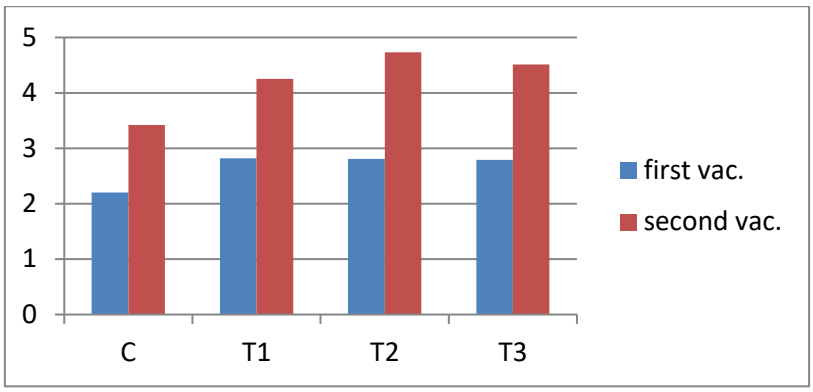

Fig 1- The volumetric standard rate of agglutination-inhibiting Newcastle agglutinin antibodies was determined after the first and second vaccinations.

The statistical analysis test results showed that the treated groups (T1, T2, T3) were superior to the control group in terms of volumetric standard rate of Newcastle antibodies measured by ELISA after the first vaccination, while the two groups (T2, T3) were significantly superior to the first treatment group (T1) with a significant difference. It has a P value of 0.05.
The statistical analysis revealed that the treatment groups (T1, T2, and T3) were significantly higher than the control group (C) after the second vaccination, with a significant difference (P < 0.05). It was also discovered that the four groups performed significantly better after the second immunization than they did after the first.

Table. (2) ELISA volumetric Newcastle antibody titer rate test revealed Following first and second vaccination.

group	standard antibodies Log ^{10} First vac.	standard antibodies Log ^{10} Second vac.
C	2.78 ± 0.19 Bb	3.2 5 ± 0.7 Ba
T1	3.39 ± 0.15 Acb	4.8 2 ± 0.05 Aa
T2	4.02 ± 0.16 Ab	4.93 ± 0.06 Aa
T3	4.21 ± 0.35 Ab	4.89 ±0.03 Aa

* The numbers represent averages and standard deviations. * Capital letters represent the vertical statistical reading (between totals), while lowercase letters represent the horizontal statistical reading (between times). The similar letters indicate that no statistically significant differences exist between the transactions, whereas the different letters indicate that significant differences exist at the level of probability. (P < 0.05).

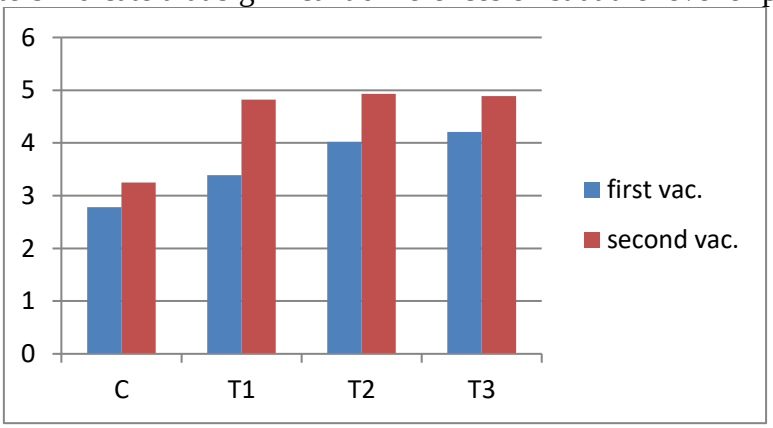

Fig 2- The volumetric titer of the Newcastle antibody ELISA test revealed After the first and second vaccinations.

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Table No. (3) The effect of cinnamon on the average number of white blood cells for the treatment groups is shown here. When compared to the negative control group, the statistical analysis revealed a significant increase in white blood cells in groups T1, T2, and T3.

Table. (3) Effect of Cinnamon on the WBCs (10^3 /mm^3) after first to second vaccination

group	First vac. WBCs (10^3 /mm^3)	Second vac. WBCs (10^3 /mm^3)
C	13.9 ± 0.19 Bb	14.21 ± 0.13 Ba
T1	19.1 ± 0.21 Ab	25.12 ± 0.35 Aa
T2	18.91 ± 0.07 Ab	22.1 1 ± 0.22 Aa
T3	18.8 ± 0.34 Ab	20.89 ± 0.45 Aa

* The numbers represent averages and standard deviations. * Capital letters represent the vertical statistical reading (between totals), while lowercase letters represent the horizontal statistical reading (between times). The similar letters indicate that no statistically significant differences exist between the transactions, whereas the different letters indicate that significant differences exist at the level of probability. ($P < 0.05$).

Table No. (4) shows the effect of cinnamon on the heterophil/lymphocyt (H/L) rate for each experiment group.

The statistical analysis revealed that there was a significant decrease in this percentage in the treatment groups compared to the control group, and there were no significant differences between the treatment groups after the first and second vaccines.

Table (4) The effect of adding cinnamon on the ratio of heterophil/lymphocyt (H /L) after first to second vaccination

Group	First vac.	Second vac.
C	0.5 2 ± 0.03 Aa	0.4 1 ± 0.02 Ab
T1	0.43 ± 0.02 Ba	0.29 ± 0.03 Bb
T2	0.39 ± 0.02 Ba	0.21 ± 0.008 Bb
T3	0.41 ± 0.02 Ba	0.2 8 ± 0.008 Bb

* The numbers represent averages and standard deviations. * Capital letters represent the vertical statistical reading (between totals), while lowercase letters represent the horizontal statistical reading (between times). The similar letters indicate that no statistically significant differences exist between the transactions, whereas the different letters indicate that significant differences exist at the level of probability. ($P < 0.05$).

Discussion

Many studies have been conducted on the possibility of using a vaccine to reduce the severity of an important disease, such as Newcastle disease, which can cause massive economic losses, and then improving the immune performance of that vaccine and increasing white blood cell counts. Plant extracts, such as cinnamon, are one of the most commonly used methods (8). T-cells are classified as effector or helper T-cells based on their function, and the T-cell response includes the production of interferon-gamma and cytokines that activate phagocytic cells as their ability to phagocytize and kill pathogens increases. representing T cells' ferocious arm (13,14). Phagocytic heterophil cells in birds produce IL-1, IL-6, and IL-8 (15). Cinnamon's role in stimulating the production of (IL-1) and (IL-6), as well as its role in macrophage activation of Th2 cells and B-cell activation for further differentiation into plasma cells, could explain the high levels of agglutination-inhibiting antibodies found in Cinnamon-treated chick serum.

Immunological memory, as memory T cells develop and produce antibodies, could explain the significant increase in antibodies in the second immunization compared to the first. The study (16) looked at CD4+ T cells in the same host after primary and secondary infections and discovered that they respond quickly when the pathogen is discovered later (17). Cinnamon may increase thymus, spleen, and Fabricius bursa weights

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by stimulating the bursa and thymus glands' production of immunoglobulins IgG and IgM. This is consistent with the findings of (18), who supplemented chicks with cinnamon powder at levels of 3, 5, and 7%, and (4), who discovered similar results using 300 mg/kg cinnamon oil.

When the treatment groups were compared to the positive and negative control groups, the number of WBCs (103/mm3) increased significantly. It could be due to immune system stimulation and the production of different types of white blood cells (19), and these findings are consistent with the findings of the two researchers (20).

The results were particularly favorable for lymphocytes, which were associated with an increase in the rates of immunoglobulins, particularly beta and gamma types, and this finding was supported by (21), who discovered a significant increase in the number of white blood cells in the cinnamon-treated groups compared to the negative control group. The H/L cell ratio is regarded as a global indicator of true health status, as an increase in this ratio indicates the presence of stress (22). A decrease in this ratio, on the other hand, indicates good breeding health and the absence of any stressors for the bird, as stress is effective. It stimulates the adrenal gland and increases the secretion of special hormones like corticosteroid hormone, which has a direct effect on the decomposition of lymphocytes produced from lymphoid tissues and the spleen (23), resulting in an increase in heterogeneous cells and, as a result, an increase in L/H. (24).

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